L14 ANSWER 2 OF 7 MEDLINE
AN 92061510 MEDLINE
DN 92061510 PubMed ID: 1953303
TI Intracellular serine protease-4, a new intracellular serine protease activity from **Bacillus subtilis**.
AU Sheehan S M; Switzer R L
CS Department of Biochemistry, University of Illinois, Urbana 61801.
NC AI11121 (NIAID)
SO ARCHIVES OF MICROBIOLOGY, (1991) 156 (3) 186-91.
Journal code: 7YN; 0410427. ISSN: 0302-8933.
CY GERMANY: Germany, Federal Republic of
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 199112
ED Entered STN: 19920124
Last Updated on STN: 20000303
Entered Medline: 19911217
AB A previously undiscovered intracellular serine protease activity, which we have called intracellular serine protease-4, was identified in extracts of stationary **Bacillus subtilis** cells, purified 260 fold from the cytoplasmic fraction, and characterized. The new protease was stable and active in the absence of Ca2+ ions and hydrolyzed azocasein and the chromogenic substrate carbobenzoxy-carbonyl-alanyl-alanyl-leucyl-p-nitroanilide, but not azocollagen or a variety of other chromogenic substrates. The protease was strongly inhibited by phenylmethylsulfonylfluoride, chymostatin and antipain, but not by chelators, sulfhydryl-reactive agents or trypsin inhibitors. Its activity was stimulated by Ca2+ ions and gramicidin S; its pH and temperature optima were 9.0 and 37 degrees C, respectively. Although intracellular serine protease-4 was immunochemically distinct from intracellular **serine protease-1**, it was absent from a mutant in which the **gene** encoding the latter was disrupted.

L14 ANSWER 1 OF 7 MEDLINE
AN 95291100 MEDLINE
DN 95291100 PubMed ID: 7773103
TI A new alkaline serine protease from alkalophilic **Bacillus** sp.: cloning, sequencing, and characterization of an intracellular protease.
AU Yamagata Y; Ichishima E
CS Department of Applied Biological Chemistry, Faculty of Agriculture, Tohoku University, Sendai, Japan.
SO CURRENT MICROBIOLOGY, (1995 Jun) 30 (6) 357-66.
Journal code: BMW; 7808448. ISSN: 0343-8651.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS B
OS GENBANK-D37921
EM 199507
ED Entered STN: 19950720
Last Updated on STN: 20000303
Entered Medline: 19950713
AB To obtain a new serine protease from alkalophilic **Bacillus** sp. NKS-21, shotgun cloning was carried out. As a result, a new protease **gene** was obtained. It encoded an intracellular **serine protease** (ISP-**1**) in which there was no signal sequence. The molecular weight was 34,624. The protease showed about 50% homology with those of intracellular **serine proteases** (ISP-**1**) from **Bacillus subtilis**, B. polymyxa, and alkalophilic **Bacillus** sp. No. 221. The amino acid residues that form the catalytic triad, Ser, His and Asp, were completely conserved in comparison with subtilisins (the extracellular proteases from **Bacillus**). The cloned intracellular protease was expressed in Escherichia coli, and its purification and characterization were carried out. The enzyme showed stability under alkaline condition at pH 10 and tolerance to surfactants. The cloned ISP-1 digested well nucleoproteins, clupein and salmin, for the substrates.

L14 ANSWER 3 OF 7 MEDLINE
AN 89053875 MEDLINE
DN 89053875 PubMed ID: 3142851
TI **Gene** encoding a minor extracellular protease in **Bacillus subtilis**.
AU Sloma A; Ally A; Ally D; Pero J
CS BioTechnica International, Inc., Cambridge, Massachusetts 02140.
SO JOURNAL OF BACTERIOLOGY, (1988 Dec) 170 (12) 5557-63.
Journal code: HH3; 2985120R. ISSN: 0021-9193.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
OS GENBANK-M22407
EM 198901
ED Entered STN: 19900308
Last Updated on STN: 19900308
Entered Medline: 19890105
AB The **gene** for a minor, extracellular protease has been identified in **Bacillus subtilis**. The **gene** (epr) encoded a primary product of 645 amino acids that was partially homologous to both subtilisin (Apr) and the major internal **serine protease** (ISP-**1**) of B. **subtilis**. Deletion analysis indicated that the C-terminal 240 amino acids of Epr were not necessary for activity. This C-terminal region exhibited several unusual features, including a high abundance of lysine residues and the presence of a partially homologous sequence of 44 amino acids that was directly repeated five times. The epr **gene** mapped near sacA and was not required for growth or sporulation.

L14 ANSWER 4 OF 7 MEDLINE
AN 88086859 MEDLINE
DN 88086859 PubMed ID: 3121583
TI Control of intracellular serine protease expression in **Bacillus subtilis**.
AU Ruppen M E; Van Alstine G L; Band L
CS Fermentation Research and Development, Genencor, Inc., South San Francisco, California.
SO JOURNAL OF BACTERIOLOGY, (1988 Jan) 170 (1) 136-40.
Journal code: HH3; 2985120R. ISSN: 0021-9193.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 198802
ED Entered STN: 19900305
Last Updated on STN: 20000303
Entered Medline: 19880210
AB Expression of the major intracellular **serine protease** (ISP-**1**) **gene** of **Bacillus subtilis** was studied by using a translational fusion plasmid in which the isp promoter region was fused to the lacZ **gene**. beta-Galactosidase activity, used to measure transcription from the isp promoter, was produced immediately after the end of exponential growth, whereas intracellular protease activity was not detected until 4 h later. These results are consistent with a previous suggestion that ISP-1 initially accumulates in the cell in an enzymatically inactive form. ISP-1 activity was detected in all of the sporulation-deficient strains examined, and the amount of protease activity always corresponded to the amount of beta-galactosidase activity. These results indicate that the activation of ISP-1 is not dependent on a sporulation-specific **gene** product. Expression of ISP-1 is regulated by a number of mutations known to affect the expression of extracellular enzymes. In sacU(h) and sacQ(h) mutants, the expression of ISP-1 was 10-fold higher than in the wild-type strain. In catA, hpr, and scoC strains, expression of ISP was stimulated two- to threefold, whereas in sacU mutants the expression of ISP-1 was reduced to less than 10% of the wild-type level. The temporal expression and activation of ISP-1 was not affected by any of these mutations. This is the first evidence that the expression of a native intracellular protein is affected by these hyperproduction mutations.

L14 ANSWER 5 OF 7 MEDLINE
AN 87083409 MEDLINE
DN 87083409 PubMed ID: 3098735
TI Construction and properties of an intracellular serine protease mutant of **Bacillus subtilis**.
AU Band L; Henner D J; Ruppen M
SO JOURNAL OF BACTERIOLOGY, (1987 Jan) 169 (1) 444-6.
Journal code: HH3; 2985120R. ISSN: 0021-9193.
CY United States
DT Journal; Article; (JOURNAL ARTICLE)
LA English
FS Priority Journals
EM 198702
ED Entered STN: 19900302
Last Updated on STN: 20000303
Entered Medline: 19870206
AB An intracellular **serine protease** (ISP-**1**) mutant of **Bacillus subtilis** was created by introducing a frameshift into the coding region of the cloned **gene**. Intracellular protease activity in the mutant was very low, yet sporulation in both nutrient broth and minimal medium was normal. The rate of bulk protein turnover in the mutant was slightly slower than that in the wild-type strain. These results suggest that the **gene** for ISP-1 is not essential and that ISP-1 is not the major enzyme involved in protein turnover during sporulation.